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UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF
PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES

TXR No.: 0052869

MEMORANDUM

DATE: September 22, 2004

SUBJECT: **CAPTAN:** Fourth Report of the Cancer Assessment Review Committee

PC Code: 081301

FROM: Jessica Kidwell, Executive Secretary
Cancer Assessment Review Committee
Health Effects Division (7509C)

TO: William Burnam, Sr. Science Advisor (IO)
Ray Kent, Branch Chief (RRB4)
Health Effects Division (7509C)

Susan Jennings (RRB3)
Michael Goodis, Branch Chief (RRB3)
Cathryn O'Connell (RRB2)
Special Review and Reregistration Branch (SRRD) (7508C)

The Cancer Assessment Review Committee met on June 9, 2004 to re-evaluate the carcinogenic potential of Captan. Attached please find the Final Cancer Assessment Document.

cc: J. Pletcher
Y. Woo

CANCER ASSESSMENT DOCUMENT

FOURTH EVALUATION OF THE CARCINOGENIC POTENTIAL OF **CAPTAN** PC CODE 081301

FINAL REPORT

September 22, 2004

CANCER **A**SSESSMENT **R**EVIEW **C**OMMITTEE
HEALTH EFFECTS DIVISION
OFFICE OF PESTICIDE PROGRAMS

DATA PRESENTATION: William Burnam

DOCUMENT PREPARATION: Jessica Kidwell, Executive Secretary

COMMITTEE MEMBERS IN ATTENDANCE: (Signature indicates concurrence with the assessment unless otherwise stated).

Karl Baetcke	
Lori Brunsman, Statistician	On Maternity Leave
William Burnam, Chair	
Marion Copley	
Vicki Dellarco	
Abdallah Khasawinah	Minority Opinion Attached
Nancy McCarroll	
Tim McMahon	
Esther Rinde	Minority Opinion Attached
Jess Rowland	
Linda Taylor	

NON-COMMITTEE MEMBER IN ATTENDANCE (Signature indicates concurrence with the pathology report)

John Pletcher, Consulting Pathologist — See attached sheet

OTHER ATTENDEES: Susan Jennings (conference call) (SRRD/RRB3), Yong-Hwa Kim (Visiting Scholar HED), Ray Kent (HED/RRB4), Alberto Protzel (HED/TB), Michael Goodis (SRRD/RRB3)

CAPTAN

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I. INTRODUCTION

On June 9, 2004 the Cancer Assessment Review Committee of the Health Effects Division of the Office of Pesticide Programs met to re-evaluate the carcinogenic potential of Captan.

II. BACKGROUND INFORMATION

Captan (N-trichloromethylthio-4-cyclohexene-1,2-dicarboximide) is a fungicide registered by Makhteshim-Agan and Arvesta Corporation for the control of fungal diseases in crops.

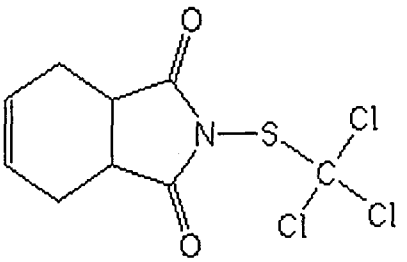

Figure 2 - Captan

Captan was classified as a B2 probable human carcinogen by the EPA and a linear low dose (Q1*) risk assessment based on adenomas and adenocarcinomas in the small intestine of both sexes of mice was recommended (US EPA, 1986, 1988a).

In 2001 the registrants of captan (Captan Task Force or CTF) requested that EPA re-evaluate the cancer classification of Captan. EPA agreed to a re-evaluation of captan's cancer classification managed by an independent Third Party. On September 3-4, 2003, an independent Third Party of outside experts in various fields and affiliations, recruited and managed by TERA (Toxicology Excellence for Risk Assessment, Cincinnati, Ohio) reviewed the captan cancer mode of action data. This panel concluded that captan acted through a non-genotoxic threshold mode of action.

In 2004, the CTF submitted the results of the Independent Expert Panel meeting to the EPA for review. The report entitled "Analysis of the Appropriate Cancer Classification of Captan under EPA's Current Guideline" dated April 9, 2004 (MRID 46247701) is the result of the CTF's incorporation of comments from the third party review. The report entitled "Scientific Analysis of the Data Relating to the Reclassification of Captan under EPA's New Guidelines for Carcinogen Risk Assessment" by Wilkinson et al. 2004 was the subject of a September 3-4, 2003 meeting by this group of outside expert peer reviewers. Their comments on this draft and a later draft were incorporated to form the basis for this current "Analysis" dated April 9, 2004. It is this final "Analysis" that would be the basis for EPA's decision of whether or not to accept the proposed cancer mode of action rationale.

III. MATERIALS REVIEWED

EPA's evaluation of an independent third party review was a new approach intended to save both time and resources. The CARC's task was to review the information and determine if they can agree with the conclusions of the third party peer review group regarding the mode of action of captan. The following critical materials were reviewed for the CARC meeting on June 9, 2004.

1. Strawson, J. 2004. Report of Peer Review Meeting Cancer Assessment for Captan. September 3-4, 2003. Project Number: CTF/0204. Final Meeting Report Date: November 21, 2003. MRID 46196301.

2. Wilkinson, C., Arce, G., Gordon, E. 2004a. Scientific Analysis of the Data Relating To The Reclassification of Captan Under EPA's New Guidelines for Carcinogen Risk Assessment. Project Number: CTF/0104. Report Date: January 13, 2004. Unpublished study prepared by Makhteshim-Agan of North America Inc. MRID No. 46173001.

3. *TERA.* 2004. Peer Review Comments on Revised Captan Report.

4. Wilkinson, C., Arce, G., Gordon, E. 2004b. Analysis of the Appropriate Cancer Classification of Captan under EPA's Current Guidelines. Project Number: CTF 0304 Report Date: April 9, 2004. Unpublished study prepared by Captan Task Force. MRID 46247701.

5. U.S. EPA. 1986. Peer Review of Captan. December 29, 1986. Carcinogenicity Peer Review Committee, US. Environmental Protection Agency, Washington, DC.

6. U.S. EPA. 1988a. Second Peer Review of Captan, Addendum. July 20, 1988. Carcinogenicity Peer Review Committee, US. Environmental Protection Agency, Washington, DC.

7. U.S. EPA. 1988b. *Ad Hoc* Committee Meeting on Captan. September 14, 1998. Carcinogenicity Peer Review Committee,, US. Environmental Protection Agency, Washington, DC.

8. Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) Scientific Advisory Panel. 1985. Review of a Set of Scientific Issues being Considered by EPA in Connection with the Special Review on Captan. Meeting Date September 26, 1985. Report Date: October 4, 1985.

IV. WEIGHT-OF-EVIDENCE CONSIDERATION

A. HAZARD IDENTIFICATION

Mouse

- **The weight of evidence indicates that captan's carcinogenicity is limited to a single tumor type (adenomas and adenocarcinomas in the small intestine, primarily the proximal portion of the duodenum) in both sexes of a single species (mouse).** The small intestine tumors were seen in three carcinogenicity studies (NCI, 1977[1]; Wong et al. 1981[2]; Daly and Knezevich, 1983[3]). Tumors were observed in females at dietary dose levels of at least 800 ppm (~120 mg/kg/day) and in males at dose levels of at least 6000 ppm (~900 mg/kg/day).

Rat

- Following review and discussion at the June 9, 2004 CARC meeting of the Captan Task Force Paper and comments from the Third Party reviewers, the CARC agreed with the CTF's bottom line conclusion that the results of the rat bioassays provide no evidence that captan is associated with kidney tumors in male rats or uterine tumors in female rats, and, therefore, these tumors do not add to the weight-of-evidence considerations for the carcinogenicity of captan.

  a) The CARC did not consider the **kidney** tumors in Charles River CD rats to be treatment-related. The CARC was in general agreement with the conclusions of the Captan Task Force. The CARC's conclusions were based on the following: 1) the finding of increased kidney adenomas was seen in only one of three rat carcinogenicity studies at adequate dose levels (Goldenthal et al. 1982[4], US EPA, 1986); 2) the increase was seen in male rats only; 3) EPA concluded that there was a borderline statistical increase in combined kidney tumors in male rats (trend, no pairwise) that was slightly

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[1]NCI. 1977. Bioassay of captan for possible carcinogenicity. National Cancer Institute, Carcinogenesis Technical Report Series Nol 15, CAS No. 133-06-2. DHEW Publication No. (NIH) 77-815. Bethesda, Maryland. MRID 00060436.

[2]Wong, ZA, Bradfield, LG, Atkins, BJ. 1981. Lifetime oncogenic feeding study of captan technical (SX-944) in CD-1 mice (ICR derived). Chevron Environmental Health Center, Report Number:Socal 1150. Chevron Chemical Company. MRID 00068076.

[3]Daly, IW, Knezevich, A. 1983. A lifetime oral oncogenicity study of captan in mice. BioDynamic Laboratories, Report Number:80-2491. Chevron Chemical Company. April 6, 1983. MRID 00126845 .

[4]Goldenthal, E. Warner, M, and Rajasekaran, D. 1982. Two year oral toxicity/carcingenicity study of captan in rats. International Research and Development Corp, Report No. 153-097. Stauffer Chemical Company, Richmond, CA. MRID 00120316.

outside the historical control range; 4) the FIFRA SAP indicated that the "rat studies are equivocal at best in indicating oncogenicity" (FIFRA SAP, 1985); 5) there was no increase in focal preneoplastic lesions usually associated with renal tubular tumors; 6) given the high reactivity of captan and its metabolite, thiophosgene, it is unlikely that they could cause formation of tumors at remote sites.

b) The CARC questioned whether or not the **uterine** tumors in Wistar rats were treatment-related and concluded that they did not contribute to the overall weight-of-evidence for carcinogenicity classification. In addition, the CARC was in general agreement with the conclusions of the Captan Task Force. The CARC's conclusions were based on the following considerations: 1) the finding of increased uterine tumors was seen in only one of three rat carcinogenicity studies at adequate dose levels (Til et al. 1983[5], US EPA, 1986); 2) the increased uterine tumors were statistically significantly increased at the high dose, however, because the study was 120 weeks in duration, there are no appropriate historical control data available for comparison; 3) given the high reactivity of captan and its metabolite, thiophosgene, it is unlikely that they could cause formation of tumors at remote sites.

B. MUTAGENICITY

Captan is a well established mutagen in bacteria and to a lesser extent in cultured mammalian cells. It induces chromosome aberrations, sister chromatid exchanges and single strand DNA breaks in mammalian cells. In all *in vitro* test systems, genotoxicity is confined to the nonactivated portion of mutagenicity studies and activity is either completely abolished or markedly reduced in the presence of rat liver S9, human or rat blood, cysteine, glutathione, or other thiols. In contrast, captan is negative in 7 acceptable *in vivo* assays (1 host mediated, 1UDS, 1 bone marrow chromosome aberration, 1 micronucleus, 2 dominant lethal and 1 mouse specific locus assays) that have been submitted to the Agency, or from 2 additional bone marrow cytogenetic assays, 4 micronucleus assays, and 1 dominant lethal test either found in the open literature or a series of 4 in vivo DNA binding studies sponsored by the registrant, 3 of which are deemed inconclusive by the Agency. Thus, the number of unequivocal negative *in vivo* studies totals 14. However, these negative *in vivo* findings are contrasted by the findings of Feng and Lin (1987)[6] who reported dose-related ≥2-fold increases in micronuclei (2.7 to 5.2-fold ↑) at levels of 100 to 800 mg/kg, respectively, and dose- related and significant ($p<0.01$) increases in chromosome aberrations at levels of 600 to 1000 mg/kg in mouse bone marrow. Additionally,

[5]Til, HP, Kuper, CF and Folke, HE. 1983. Life-span carcinogneicity study of Merpan (captan). Civo Institutes:TNO Report No. V83.233/200153. MRID 00161230.

[6]Feng, JY, Lin, BY. 1987. Cytogenetic effects of an agricultural antibiotic, captan, on mouse bone marrow and testicular cells. Environ. Res. 43:359-63.

they reported a significant ($p<0.01$) increase in chromosome aberrations (fragmentation and translocations) in spermatogonia at 800 mg/kg and in primary spermatocytes at 1000 mg/kg (fragmentation and translocation). The Sponsor's representative stated that these results were questionable because of certain mathematical errors and difficulty in data interpretation, plus, the purity and source of the test sample were of concern. While the scientific merits and findings of the study should be viewed with caution, the lack of confirmation of the findings renders them in conflict with the sizable negative *in vivo* database. Furthermore, the evidence of chromosome damage in germinal cells is in disagreement with the 3 negative dominant lethal assays, the negative EPA-sponsored heritable translocation study in mice and the negative mouse specific locus assay.

There is, however, concern regarding the claims of the Sponsor's representative that "there is no evidence of captan DNA binding." This position was based on the "negative" or "no conclusion" from a series of *in vivo* DNA binding studies sponsored by a pesticide company. One of these studies and portions of a second assay were submitted to the Agency and found to be unacceptable for regulatory purposes because no conclusions could be reached due to technical problems and/or unexpected results with known test agents.

In a memorandum dated June 9, 1998, Yin-tak Woo, Senior Toxicologist from OPPTS stated:

" The key argument that Captan is not genotoxic *in vivo* because of the apparent lack of DNA binding does not appear to be scientifically sound because the registrant has not investigated the possible crosslinking activity expected from thiophosgene."

He goes on to say that because of rapid detoxification, thiophosgene produced in the duodenum is unlikely to reach other target cells and, therefore, would be expected to yield negative *in vivo* genotoxicity data if the target cells are outside the duodenum. In response to these comments, the data from the study of Chidiac and Goldberg (1987)[7] demonstrated that captan administered either in the diet or by oral gavage at doses higher than the tumorigenic levels and also as single administrations representing ½ of the LD_{50} or as cumulative doses exceeding the LD_{50} did not induce nuclear aberrations (i.e.,apoptotic bodies and micronuclei) in the proximal small intestine of CD-1 mice. Furthermore, captan was not found to induce nuclear abnormalities in crypt cells of the epithelium of the small intestine of CD-1 mice receiving various samples with different degrees of captan purity; 2 major captan impurities [1,2,3,,6-tetrahydrophthalimide and bis(trichloromethyl) disulfide] were also negative. Similarly, pretreatment of the mice with an inhibitor of glutathione biosynthesis (L-buthionine-S-, R-sulfoximine) also failed to produce positive results.

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[7]Chidiac, P. Goldberg, MT. 1987. Lack of induction of nuclear aberrations by captan in mouse duodenum. Environ. Mut. 9:297-306. MRID 43405103.

Overall, the data support the contention that captan is an *in vitro* mutagen and clastogen that is not active in the whole animal because it reacts with thiols or proteins that rapidly deactivate captan or its reactive breakdown product (thiophosgene). It is, therefore, likely that captan and thiophosgene will undergo breakdown either by hydrolysis or reaction with thiols before both substances have access to stem cells deep within the duodenal crypts. It is also probable, given the rapid half-live of captan (<1 second) or thiophosgene (0.6 seconds) in blood, that neither substance can reach tissues distant from the portal of entry to cause DNA damage.

The CARC concluded, therefore, that captan is not genotoxic *in vivo*, and the weight-of-the-evidence supports a nongenotoxic mode of action for captan.

C. PROPOSED MODE OF ACTION

The CARC accepts the proposed mode of action as set forth by the CTF that suggests that "captan induces adenomas and adenocarcinomas in the duodenum of the mouse by a non-genotoxic mode of action involving cytotoxicity and regenerative cell hyperplasia that exhibits a clear dose threshold. These responses are reversible following cessation of captan exposure. There is a strong causal association (dose-response, temporality) indicating that tumor formation is secondary to cytotoxicity and hyperplasia and that the latter is a key event in the sequential cascade of events leading to cancer". As stated in the CTF document, the proposed sequence of events (irritation→inflammation/cytotoxicity→cell proliferation→tumors) in the carcinogenic process is as follows:

"1. Following oral ingestion, captan is rapidly degraded to THPI, thiophosgene, and other reactive species in the stomach and the proximal part of the small intestine-breakdown occurs by either hydrolysis or reaction with GSH and other thiols;
2. Captan and thiophosgene, both strong chemical irritants, cause inflammation, cytotoxicity, and necrosis of the epithelial cells of the villi in the proximal portion of the duodenum;
3. Cytoxicity causes cells to be sloughed off the tips of the villi at a faster rate than normal, resulting in a shortening of the height of the villi;
4. The enhanced cell loss in the villi causes an increase in crypt cell proliferation and regenerative hyperplasia in the stem cells from which the epithelial cells are derived;
5. Prolonged hyperplasia in the stem cells overwhelms their capacity to repair damaged DNA and increass the probability of cloning a transformed cell; and
6. The increased cloning of cells containing naturally occuring DNA damage leads to an increased incidence of duodenal adenomas and adenocarcinomas."

Currently, no data were presented at this time to suggest a different mode of action in children. The reactivity of captan and thiophosgene is unlikely to depend on an activation system and, therefore, no difference in susceptibility between adults and children is expected.]

D. STRUCTURE ACTIVITY RELATIONSHIP (SAR)

Captan is structurally related to folpet. Folpet and captan have the same side chain (which may convert to thiophosgene, a highly reactive compound). Both captan and folpet are associated with an increased incidence of intestinal tumors in the CD-1 mouse following dietary exposure. Folpet is classified as a Group B2-Probable Human Carcinogen based on intestinal tumors in mice.

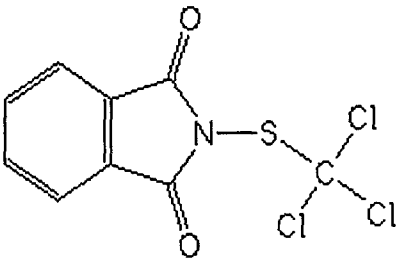


Figure 3 - Folpet

V. CLASSIFICATION OF CARCINOGENIC POTENTIAL

In accordance with the EPA Proposed *Guidelines for Carcinogen Risk Assessment* (February, 2003), the Committee classified captan as **"Likely to be carcinogenic to humans following prolonged, high-level exposures causing cytotoxicity and regenerative cell hyperplasia in the proximal region of the small intestine (oral exposure) or the respiratory tract (inhalation exposure), but not likely to be a human carcinogen at dose levels that do not cause cytotoxicity and regenerative cell hyperplasia."** In addition, captan is "Not likely to be carcinogenic to humans via dermal exposure route" due to the very low dermal penetration of captan.

This decision was based on the following weight-of-evidence considerations:

(i) The occurrence of adenomas and adenocarcinomas in the small intestine of male and female mice;

(ii) Captan is not likely to be genotoxic *in vivo*;

(iii) The weight-of-evidence suggests that captan induces small intestine tumors by a nongenotoxic mode of action involving cytotoxicity and regenerative cell hyperplasia that exhibits a clear dose threshold.

VI. QUANTIFICATION

Since the CARC determined that there was a plausible MOA, captan should be regulated based on a non-linear risk assessment. A non-linear point of departure will be selected based on all the mode of action data.

Date: September 22, 2004

To: William Burnam, Chair
Cancer Assessment Review Committee (CARC)

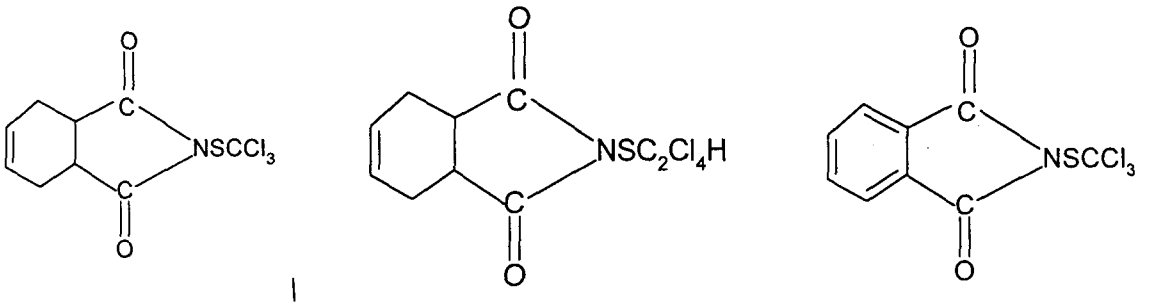
Subject: Captan Classification

From: Esther Rinde, Ph.D., D.A.B.T.
Abdallah Khasawinah, Ph.D.

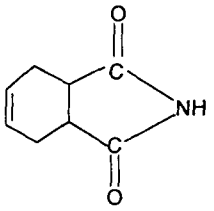
We have briefly described below why we feel the classification for captan should have been "Likely to Be Carcinogenic to Humans."

STRUCTURAL CONSIDERATIONS

The CPRC only considered folpet which has the same N-trichloro methyl thio moiety, to be a structural analog of captan, however, both captan and captafol have the same ring structure and both are initially hydrolyzed to tetrahydophthalimide (THPI). THPI may be associated with the tumors seen in the rat.



Captan Captafol Folpet



THPI

TUMORS IN THE RAT

The analog captafol was associated with an increased incidence of renal tumors in Charles River CD male rats and captan was associated with an increased incidence of renal tumors in males of the same rat strain.

- Kidney tumors are rare in rats.

- In Charles River CD rats administered captan, the incidence of combined renal adenoma/carcinoma was increased at 100 mg/kg/day and 250 mg/kg/day (4% and 5.7% vs 0 in concurrent controls); there was a positive dose-related trend and the increased incidence at the HDT was statistically significant and outside HC (0-1.7%).

- The CARC's states as a reason for dismissing the kidney tumors: "the finding of increased kidney tumors in only one of the three carcinogenicity studies"; however there were **differences in dosages and/or duration** which need to be considered.

  In the IRDC study[8] with Charles River CD rats (1982) the doses were **0, 25, 100 or 250 mg/kg/day** in a **two-year** study and kidney tumors were increased at the mid and high doses. In the NTP study with Osborne Mendel rats (1977) , there were no kidney tumors and the doses were **126 or 302.5 mg/kg/day**, but were only applied for **80 weeks** (no treatment last 33 weeks). Notably, in the Makhteshim-Agan study[9] with Wistar rats (1983) in which there were no kidney tumors, the doses were only **0, 6.25, 24, or 98 mg/kg/day** for 30 months.

- The increase in uterine fibrosarcomas (malignant) which only occurred in the 1983 Wistar rat study (8% vs 0 in concurrent controls) was outside HC[10] range. **Contrary to the Registrant's claim, the accompanying fibromatous polyps, should not be combined with the fibrosarcomas.** (Even if we *were* to combine polyps with the sarcomas, the malignant component (sarcomas) would still be considered separately.) Furthermore, uterine sarcomas are <u>rare</u> in both humans and rats.

- Thus there <u>were</u> kidney and uterine tumors associated with captan administration in the rat. No mechanistic data for either of these tumors were provided (kidney or uterine).

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[8]This study is referred to as: *Goldenthal et al. (1982)* in the CARC report.

[9]This study is referred to as: *Til et al. (1983)* by the Registrant.

[10]TXR 006043, Memo dated Feb.5, 1986: In this lab, uterine sarcomas occurred with an incidence of 0-2%; in 7 of 8 studies the incidence was zero. Four of these studies were of 30 month duration, the remaining 4 were 24-29 months.

ISSUES RELATED TO PROPOSED MOA

- According to the Registrant ".. the rapid breakdown of captan in blood (the half-life of captan is less than one second and that of triophosgene is approximately 0.6 seconds) precludes the possibility that it can be transported to other tissues in the circulation following oral or dermal administration. THPI, the relatively stable breakdown product, enters the systemic circulation."

  Even if the half-life is as short as is claimed, there will still always be some captan and its metabolite which can interact with cellular components. Furthermore:

- As Yin Tak stated in his June 9, 1998 "Critique of Mechanistic Studies of Captan":

  "While thiophosgene may be the principal proximate carcinogen of captan and folpet in the duodenum, **the potential influence on tumor formation, especially at other target organs, from the rest of the molecule (i.e.: tetrahydophthalimide (THPI) or phthalimide) cannot be ruled out."**

- As stated by the registrant, the relatively stable tetrahydophthalimide (THPI) "enters the systemic circulation" and could in fact be carried to distant sites, ie: kidney and uterus.

- If irritation/inflammation et al. is the *only* "MOA" for intestinal tumors in mouse with captan, why should the rat be different? Both the Panel and some CARC members noted that there appears to be no apparent explanation for this species difference between rats and mice and no satisfactory answers were provided by either group.

CONCLUSION

We feel that the weight of evidence for captan **should not** be characterized as - *Likely, but Not Likely at doses below which the Registrant's proposed MOA for mouse intestinal tumors is operative*- since there is at least some evidence of other tumors in the rat (kidney and uterine tumors for which there is no known mechanism or MOA). Regarding the proposed MOA, there is no satisfactory explanation as to the absence of intestinal tumors in the rat. **Therefore, a classification of "Likely To be Carcinogenic To Humans" would have been more appropriate as described above.**

13544

R178658

Chemical Name: Captan

PC Code: 081301
HED File Code: 61200 SRRD CDC
Memo Date: 9/22/2004
File ID: TX0052869
Accession #: 000-00-8016

HED Records Reference Center
11/17/2010